

TWO NEW WESTERN NEARCTIC *CULICOIDES* LATREILLE
(DIPTERA: CERATOPOGONIDAE) DESCRIBED FROM ALL STAGES

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Abstract.—The egg, larva, pupa, and adult of two new species of *Culicoides* Latreille from desert mountains in southern California and Baja, Mexico are described: *C. kettlei* and *C. vetustus*. Descriptions of immatures were made from laboratory-reared material. Rearing techniques and notes on behavior observed in the laboratory are presented.

Key Words: *Culicoides*, Ceratopogonidae, immatures, morphology

Bluetongue and epizootic hemorrhagic disease viruses have been suspected of contributing to lamb mortality in desert bighorn sheep populations in southern California (DeForge et al. 1982, Wehausen et al. 1987, Elliott et al. 1994). A comprehensive survey of potential vectors (*Culicoides* Latreille) occurring in bighorn habitats within the desert mountains of southern California (Mullens and Dada 1992a) revealed three undescribed species of *Culicoides*. The adults of one species, *Culicoides boydi* Wirth and Mullens have been previously described (Wirth and Mullens 1992). The present study describes all life-stages of these remaining two species and provides data on their biology and geographic distribution.

MATERIALS AND METHODS

Host-seeking females were collected using suction traps baited with 1 kg of dry ice. Some females collected at the Philip L. Boyd Deep Canyon Desert Research Center near Palm Desert, Riverside Co., CA, or near the town of Morongo Valley, San Bernardino Co., were given the opportunity to feed on heated, defibrinated sheep blood

through a parafilm or chick-skin membrane (Hunt 1994). Engorged females were separated from other midges in the collections while they were immobilized on a chill table. Female midges were then held at 21°C for 7 days. Some individual females would oviposit onto damp filter paper, when held overnight in a petri dish. Gravid females which did not oviposit were decapitated to induce oviposition.

Filter paper with eggs from an individual female was placed in a petri dish containing nutrient-enriched 1.5% noble agar. Eggs were held in a humid chamber and checked daily for hatch. First-instar larvae were offered several food sources, including a nutrient rich liquid diet consisting of bacteria, algae, and yeast used for rearing colonized *Culicoides variipennis sonorensis* Wirth and Jones (Jones et al. 1969). In addition, the bacterial feeding nematodes, *Pelodera* sp. and *Panagrellus redivivus* (L.), were supplied as potential prey on a biweekly basis (Mullens and Velten 1994). Larval growth and feeding behavior were observed daily and the duration of egg, larval, and pupal development recorded.

Samples of eggs from associated females

were placed into 70% EtOH. Specimens were fixed (Day et al. 1997), then critical-point dried, transferred onto stubs backed with sticky tape, sputter-coated with gold-palladium, and viewed on either a JOEL JSM-35C or Phillips XL30 scanning electron microscope.

The parental adult female was preserved in 70% EtOH and slide-mounted in balsam after Wirth and Marston (1968). Fourth-instar larvae, pupae, and adults from reared offspring of the parental female were also preserved. Offspring were mounted in Canada balsam or Hoyer's medium. Descriptions of immature stages and males of both species were made from laboratory-reared material.

The terminology of Downes and Wirth (1981) is used for adults, of Lamberson et al. (1992) and Nevill and Dyce (1994) for pupae, of Murphree and Mullen (1991) for larvae, and of Becker (1961) and Campbell and Kettle (1975) for eggs. Nomenclature agrees with Borkent and Wirth (1997) and Spinelli and Ronderos (1997).

The following measurements were made from fourth-instar larvae: total length (TL), head length (HL), head width (HW), subgenal width (SGW), mandible length (ML), width across the lateral arms of the epipharynx (LAW), total width across the paired dorsal comb sclerites of the epipharynx (DCW), caudal-segment length (CSL), caudal-segment width (CSW), length of setae 'o' (OL), and the distance between their bases.

From values listed above, the following ratios were calculated: head ratio ($HR = HL/HW$), subgenal ratio ($SGR = HW/SGW$), and caudal-segment ratio ($CSR = CSL/CSW$). Illustrations were made of the morphology and chaetotaxy of the head capsule and caudal segment. The hypostoma, epipharynx, hypopharynx, and mandible were illustrated, and thoracic pigmentation and anal papillae were drawn when appropriate.

For pupae the following structures were described and illustrated: respiratory horn,

operculum, caudal segment, and the *ad*, *dl*, *dasm*, *dpm*, *lasm*, *lpm*, and *vpm* tubercles.

The holotype, allotype, and some paratypes are deposited in the National Museum of Natural History (USNM), Smithsonian Institution, Washington, DC; paratypes, as available, will be deposited in the collections of the University of California, Riverside and the California Academy of Sciences, San Francisco.

Culicoides (Haematomyidium) kettlei
Breidenbaugh and Mullens, new species
(Figs. 1–4)

Egg.—Banana-shaped. Surface with two types of ansulae arranged in longitudinal rows (Fig. 1A). Moderately stalked ansulae present on concave surface, ridges composed of flattened papillate ansulae on convex surface (Fig. 1B). Average length = $356 \pm 17 \mu$; width = $53 \pm 2 \mu$ ($n = 11$).

Larva.—Total length = 2.79 (2.16–3.51, $n = 15$) mm. *Head capsule* (Figs. 2A–C): Light brown. Small larvae, HL = 128 (122–134, $n = 29$) μ , HW = 89 (82–102, $n = 28$) μ ; shape somewhat long and narrow, SGW = 58 (54–67, $n = 30$) μ ; HR = 1.4 (1.2–1.6, $n = 27$); SGR = 1.5 (1.4–1.8, $n = 27$). *Mandible* (Fig. 2F) medium-small, ML = 35 (22–38, $n = 29$) μ ; with pointed apex and subapical rounded prominence; base with distinct point of articulation. *Hypostoma* (Fig. 2G) rounded, entire. *Labium* small, crescent-shaped and sclerotized. *Epipharynx* (Fig. 2H): Dorsal-comb sclerites with 5–7 unequal angular teeth; moderately-wide, DCW = 11 (10–13, $n = 17$) μ . Comb 2 narrow with short, rounded teeth; comb 4 wider, with rounded teeth. LAW = 49 (43–52, $n = 30$) μ ; lateral curtains composed of short, dense, filamentous processes. *Hypopharynx* apparently poorly sclerotized, not seen. *Thoracic pigmentation* (Fig. 2I): distinct lateral pattern on thoracic segments. Prothorax with arched pattern; mesothorax and metathorax with crescent and sagittate patterns, respectively. *Caudal segment* (Figs. 2D, E): Short, CSL = 231 (189–285, $n = 32$) μ and moderately-wide, CSW = 126

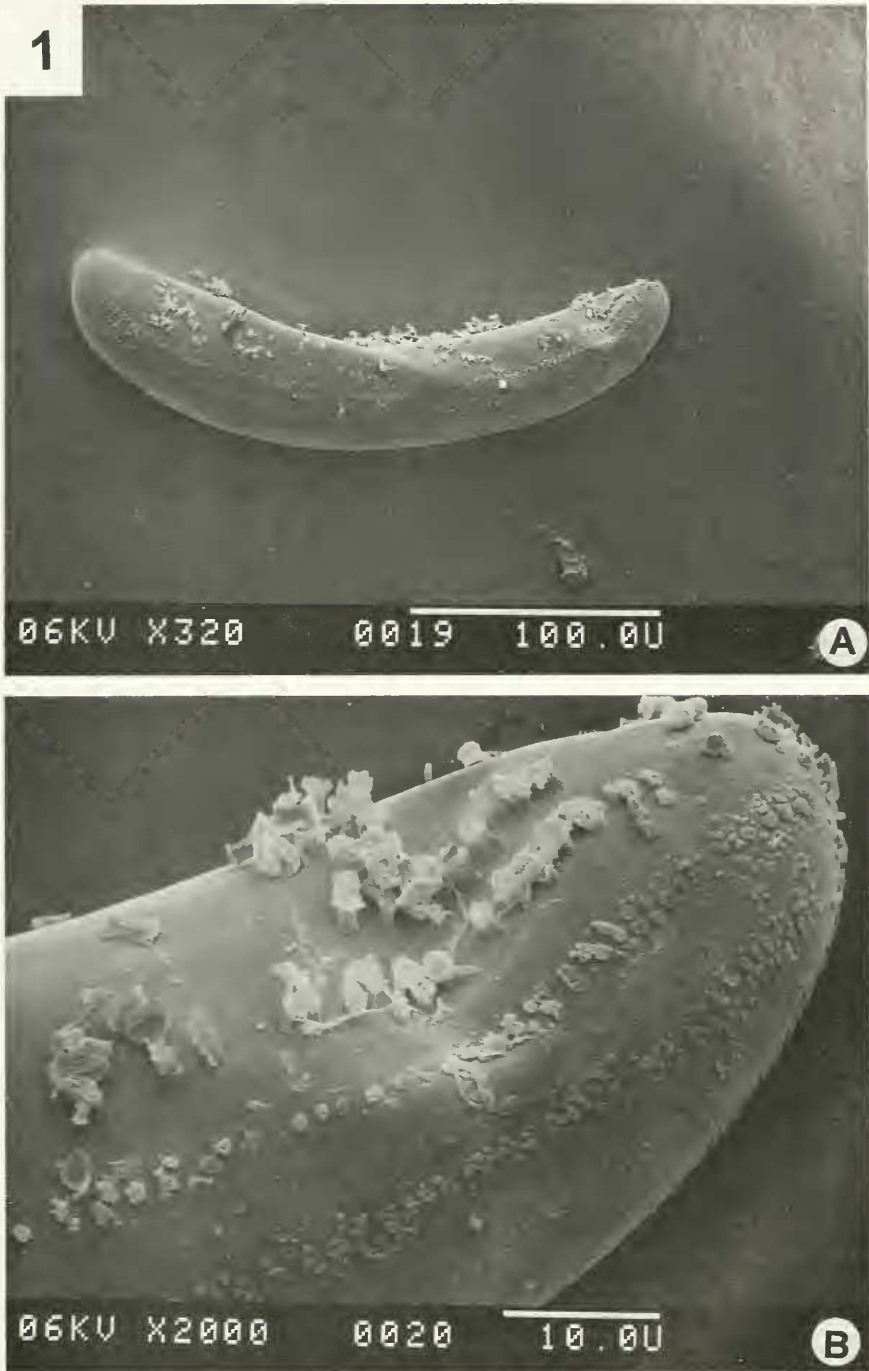


Fig. 1. *Culicoides kettlei*, egg. A, Lateral view. B, Detail of surface.

(90–179, n = 31) μ ; Oval; CSR = 1.9 (1.5–2.2, n = 31). Setae “o” short relative to the CSL, OL = 46 (38–51, n = 19) μ ; bases of

setae moderately separated, OD = 37 (26–45, n = 19) μ . Anal papillae not observed.

Material studied.—Laboratory-reared from

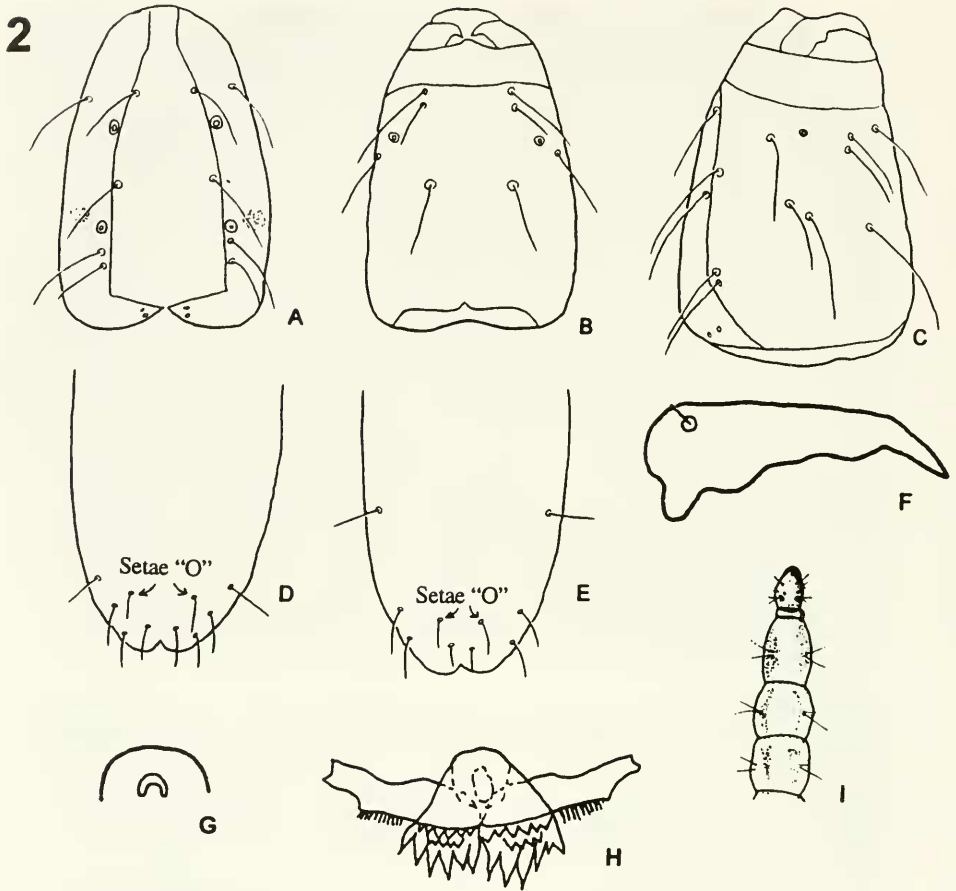


Fig. 2. *Culicoides kettlei*, larva. A–C, Head capsule. A, Dorsal view. B, Ventral view. C, Lateral view. D, Caudal segment, dorsal view. E, Caudal segment, ventral view. F, Mandible. G, Hypostoma. H, Epipharynx. I, Thoracic pigmentation, dorsal view.

eggs deposited by individual females collected in San Bernardino County, CA, Morongo Valley, 15-VI-95 (n = 3); 22-V-96 (n = 5); 14-VI-96 (n = 24), paratypes: slides 25,26,27.

Pupa.—Light yellow brown. *Respiratory horn* (Fig. 3A): yellow brown; basal half crenulated; spines, sparsely distributed. Four lateral spiracles present and 8–11 apical spiracles. Tracheal annulations associated with tracheae visible in basal portion and a reticulated pattern is visible in distal portion. *Operculum* (Fig. 3B): Yellow brown, with two types of spines, neither extending beyond the *am* tubercles. Sagittate spines occurring laterally; smaller, rounded

spines occurring medially. Anterior margin moderately notched. *Caudal segment* (Fig. 3C): Spines present at base of posterolateral processes, absent apically; band of spines along anterior margin and a patch of spines located basimedially on dorsum.

Chaetotaxy: Dorsal tubercles (Fig. 3D): 1 and 2 round with stout spine; 3 round with a short seta; 4 a slender seta; 5 a small, round pore. *ad* tubercle (Fig. 3E): spinate with two subequal spines. *dl* tubercle (Fig. 3F): with 3 unequal, slender spines. Abdomen (Fig. 3G): *dasm* tubercles: 1 with a spine and 2 a bristle. *dpm* tubercles: rounded; 1 and 2 with short spine; 3 and 4 lack setae; 5 with a short hair. *lasm* tubercle: spi-

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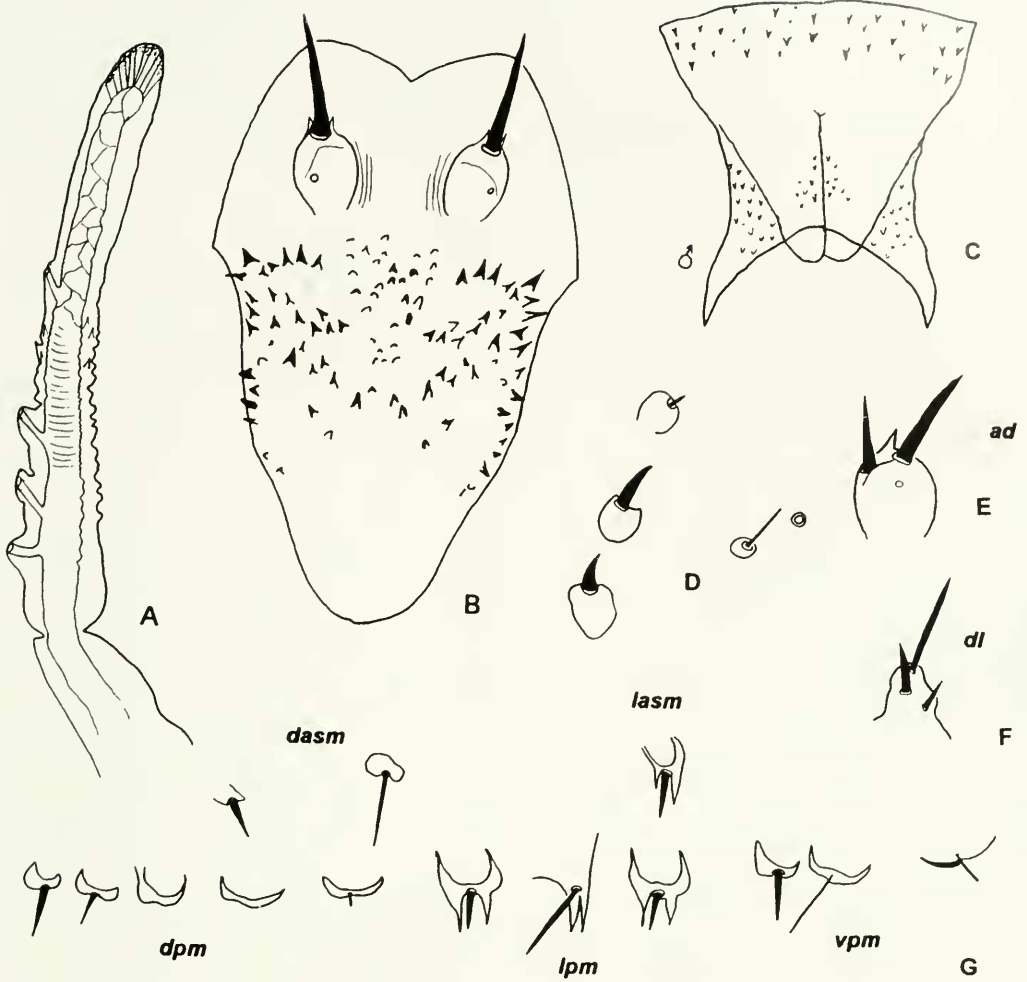


Fig. 3. *Culicoides kettlei*, pupa. A, Respiratory horn. B, Operculum. C, Caudal segment. D, Dorsal tubercles. E, ad tubercle. F, dl tubercle. G, Abdominal chaetotaxy.

nate with short spine. *lpm* tubercles: 1-3 spinate; 1 and 3 with short spine; 2 with a bristle. *vpm* tubercles: rounded; 1 with a spine; 2 and 3 with long and medium length setae, respectively.

Material studied.—Laboratory-reared from individual females collected in San Bernardino County, CA, Morongo Valley, 14-V-95 (n = 6), paratypes: slides 10,11.

Adults.—Female: Wing length 1.06 (0.92-1.19, n = 28) mm. *Head*: Eyes (Fig. 4A) bare; separated by a single facet width; without interfacetal hairs. Antenna (Fig.

4B) with lengths of flagellomeres of holotype 43-27-28-33-29-31-30-33-36-37-43-47-63 (in μ); antennal ratio 0.91 (0.74-1.04, n = 28); well-developed sensilla coeloconica present on flagellomeres 1, 4-8; 26% (7/27) of observed specimens lack sensilla on flagellomere 4. Palpus (Fig. 4C) with lengths of segments 24-46-61-20-26; palpal ratio 2.07 (1.75-2.36, n = 29); P/H ratio 0.79 (0.70-0.86, n = 18); third segment swollen with broad shallow sensory pit. *Thorax*: Brown, scutum without conspicuous pattern. Legs brown (Fig. 4D),

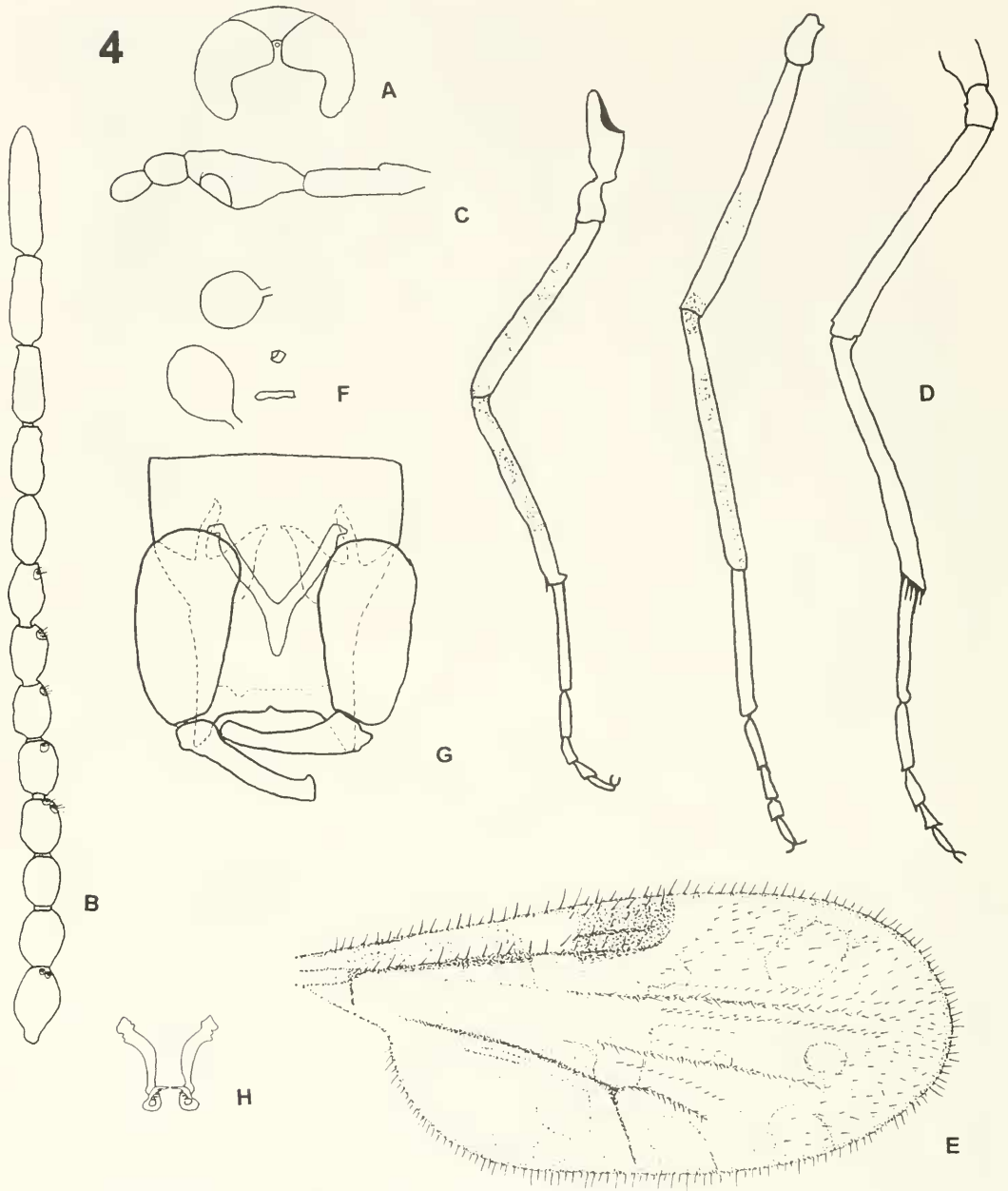


Fig. 4. *Culicoides kettlei*, adult. A-F, Female, G-H, Male. A, Eye separation. B, Antenna. C, Palpus. D, Legs, left to right, front, mid, and hind. E, Wing. F, Spermathecae. G, Genitalia, parameres omitted. H, Parameres.

femorotibial joints blackish; fore and mid femora with narrow subapical pale rings, all tibiae with narrow subbasal pale rings and hind tibiae with distal fourth pale; hind tibial comb with five spines, second from the spur longest. Wing (Fig. 4E) with a slightly

angular poststigmatic pale spot; distinct transverse pale spot present in r5; cell m1 with narrow pale spot; cell m2 with rounded pale spot located distally and with a longitudinal pale spot lying posterior to medial fork. Macrotrichia in moderate numbers

evenly distributed over wing. Halter pale. *Abdomen*: Brown. Two slightly unequal spermathecae, plus rudimentary third and sclerotized ring (Fig. 4F). Functional spermathecae with long slender necks; length, including neck, 32 (29–37, $n = 25$) μ for the larger, 29 (26–35, $n = 26$) μ for the smaller.

Male: *Genitalia* (Figs. 4G–H): Ninth sternum with scarcely perceptible caudo-median excavation, ventral membrane spiculate; apicolateral processes moderately long, pointed, caudal margin between them slightly cleft medially. Gonocoxite moderately stout; ventral root with anterior point longer, more slender than dorsal root; gonostylus moderately curved and slender with moderately bent, pointed tip. Aedeagus with basal arch extending more than two-thirds of total length; distal portion short, tapering to a simple tip. Paramere (Fig. 4H) with strong basal knob; midsection arched; distally, greatly narrowed and reflexed upon midsection, constricting to a noticeably fringed filamentous tip.

Distribution.—Southern California and northern Baja California, Mexico.

Material studied.—HOLOTYPE: ♀, Riverside County, CA, Deep Canyon, Bighorn Overlook, nr. Palm Desert, 22-IX-88 (B. A. Mullens), CO₂-baited trap. ALLOTYPE: ♂, San Bernardino County, CA, Big Morongo Canyon, Morongo Valley, 15-VI-95 (Breidenbaugh), laboratory-reared from wild-collected female. PARATYPES: CALIFORNIA. 6 ♀, same data as holotype, except 1 ♀ 24 May 1988; 6 ♀, Riverside Co., Bighorn Drive, Palm Desert, 8 October 1988, CO₂-baited trap, 1 ♀, same except 22 September 1988; 9 ♀, 4 ♂, same data as allotype except from June 1995 to July 1996 (some K. Luhring); MEXICO. 7 ♀, Mexico, Baja California, Cadavina, 19 March 1994 (C. Szijj), CO₂-baited trap.

Behavior and rearing.—In the laboratory, the average female fecundity was 74 ± 24 eggs ($n = 10$). From a single reared cohort observed daily, pupation first occurred 26 days following egg hatch. In culture, larvae

fed on *Pelodera* sp. nematodes. Adults have been collected from soil-emergence traps along a seasonal creek in the Deep Canyon watershed (Breidenbaugh and Mullens, in preparation), indicating that creek margins are used by this species as a larval development site. Feeding behavior is poorly known with a single report of an unfed female recovered from a bighorn sheep (Mullens and Dada 1992b).

Discussion.—*Culicoides kettlei*, was originally recognized by the late W. W. Wirth who referred to this species as #120 (W. W. Wirth, personal communication). Following Vargas (1960) the male genitalia fit nicely into the subgenus *Haematomyidium*. The female wing pattern is inconclusive, since the r-m crossvein is not dark. However, we feel that placement in the subgenus *Haematomyidium* is reasonable. Adult morphology is similar to that of *Culicoides (Haematomyidium) debilipalpis* Lutz, a common biting midge in the eastern U.S.. From 1985–1997 *C. debilipalpis* was synonymized with *C. lahillei* Ichès (Spinelli and Wirth 1985). Recently, however, the validity of *C. debilipalpis* was confirmed (Spinelli and Ronderos 1997). The range of *C. debilipalpis* includes the southeastern U.S. as far west as Louisiana, with a disjunct distribution that includes Costa Rica south to Argentina. In contrast, *C. lahillei* is strictly South American. The females of *C. kettlei* can be easily separated from *C. debilipalpis* and all other Nearctic members of *Haematomyidium* by the antennal sensorial pattern (1,4(5)-8) (Wirth et al. 1985). In addition, no Neotropical species of *Haematomyidium* are known to occur north of southern Mexico (Wirth et al. 1988), and their range thus does not overlap with that of *C. kettlei*.

Apart from the similarity in size, the larvae of this species are noticeably different from those of the only other North American species in this subgenus with described larvae, *Culicoides paraensis* (Goeldi) (Murphree and Mullen 1991). The hypostoma is round and smooth in this species

while lateral teeth are present in *C. paraensis*. Furthermore, the hypostoma lacks the distinct subapical notch seen in *C. paraensis*. The pupa of this species is similar to *C. paraensis* in the type and location of spines on the operculum.

Etymology.—This species is named in honor of Dr. D. S. Kettle, Emeritus Professor, Department of Entomology, University of Queensland, Australia, for his pioneering work on the biology of *Culicoides*, specifically in the discipline of the morphology of immatures.

***Culicoides vetustus* Breidenbaugh and Mullens, new species**

(Figs. 5–8)

Egg.—Cigar-shaped. Surface with flattened longitudinal rows, many not contiguous from end to end (Fig. 5A). Ansulæ flattened, not distinct, merge to form longitudinal ridges (Fig. 5B), present on all lateral surfaces, and not varying with curvature. Average length = $267 \pm 28 \mu$ and width = $49 \pm 3 \mu$ ($n = 8$).

Larva.—Total length = 3.25 (2.81–4.0, $n = 15$) mm. **Head capsule** (Figs. 6A–C): Yellow; medium sized, HL = 180 (166–192, $n = 28$) μ , HW = 123 (109–138, $n = 27$) μ , SGW = 90 (80–102, $n = 28$) μ ; overall shape long and narrow, HR = 1.5 (1.2–1.7, $n = 27$), very oblong, SGR = 1.4 (1.2–1.5, $n = 27$). Mandible (Fig. 6D) medium length, ML = 51 (48–54, $n = 24$) μ ; base wide; pointed marginal prominence basimedially, followed by a subapical notch, sharply angled to pointed apex. Hypostoma (Fig. 6E) difficult to see but rounded medially and smooth. **Epipharynx** (Fig. 6F): Dorsal-comb sclerites moderately wide, DCW = 14 (13–15, $n = 24$) μ , with 5 subequal pointed teeth/sclerite; comb 4 with many unequal pointed, rounded teeth; lateral curtains wide; teeth thin, hair-like; LAW wide relative to DCW, LAW = 60 (52–67, $n = 21$) μ , indented near lateral apex. Hypopharynx (Fig. 6G) with hypopharyngeal fringe separated into two distinct prominences by a medial notch. **Tho-**

racic pigmentation (Fig. 6H): Absent. **Caudal segment**: Short with length varying considerably among individuals, CSL = 286 (131–326, $n = 25$) μ ; narrow, CSW = 139 (86–157, $n = 26$) μ , CSR = 2.1 (1.5–2.4, $n = 25$); setae "o" of medium length, OL = 82 (61–99, $n = 18$) μ , and well separated, OD = 45 (35–53, $n = 21$) μ . Anal papillae (Fig. 6I) four deeply bifurcate pairs.

Material studied.—Laboratory-reared from eggs deposited by individual females collected in San Bernardino County, CA, Morongo Valley, 15-VI-95 ($n = 6$); 14-V-96 ($n = 15$); 14-V-96 A42 ($n = 15$), paratypes: slides 2–7.

Pupa.—Light to dark brown. **Respiratory horn** (Fig. 7A): Proximal portion light brown; roughly divided into 3 subequal portions. First section with annular tracheae; second portion annulations obscured by reticulation pattern; 3 lateral spiracular protuberances present; terminal section, dark brown with 5–9 spiracular openings apically; few scales on horn. **Operculum** (Fig. 7B): Mostly smooth, 3–8 large spines on lateral margins and patch of setae on central portion; no large spines medial to the *am* tubercles; large spines triangular. **Caudal segment** (Figs. 7C–D): Narrow V-shaped cluster of small spines on dorsal surface of both sexes. Anterior band of spines complete on both sexes; large spines on proximal $\frac{2}{3}$ of posterolateral processes; distal third, smooth dark brown.

Chaetotaxy: Dorsal tubercles (Fig. 7E): setae 1 and 2 of medium length, stout; seta 3 short, stout; seta 4 a long, slender bristle; 5 a circular pore. *ad* tubercle (Fig. 7F) with one long, one shorter setae. *dl* tubercle (Fig. 7G) with a long slender bristle and second shorter thicker bristle. Abdomen (Fig. 7H): *dasm* tubercles: setae 1 and 2 of medium thickness, 1 longer than 2. *dpm* tubercles: setae 1 and 2 equal; 3 and 4 lack setae; 5 short. *lasm* tubercle: rounded with short spine. *lpm* tubercles: spinate with shorter, stouter, equal bristles on 1 and 3, bristle 2 longer and slender. *vpm* tubercles: 2 with a

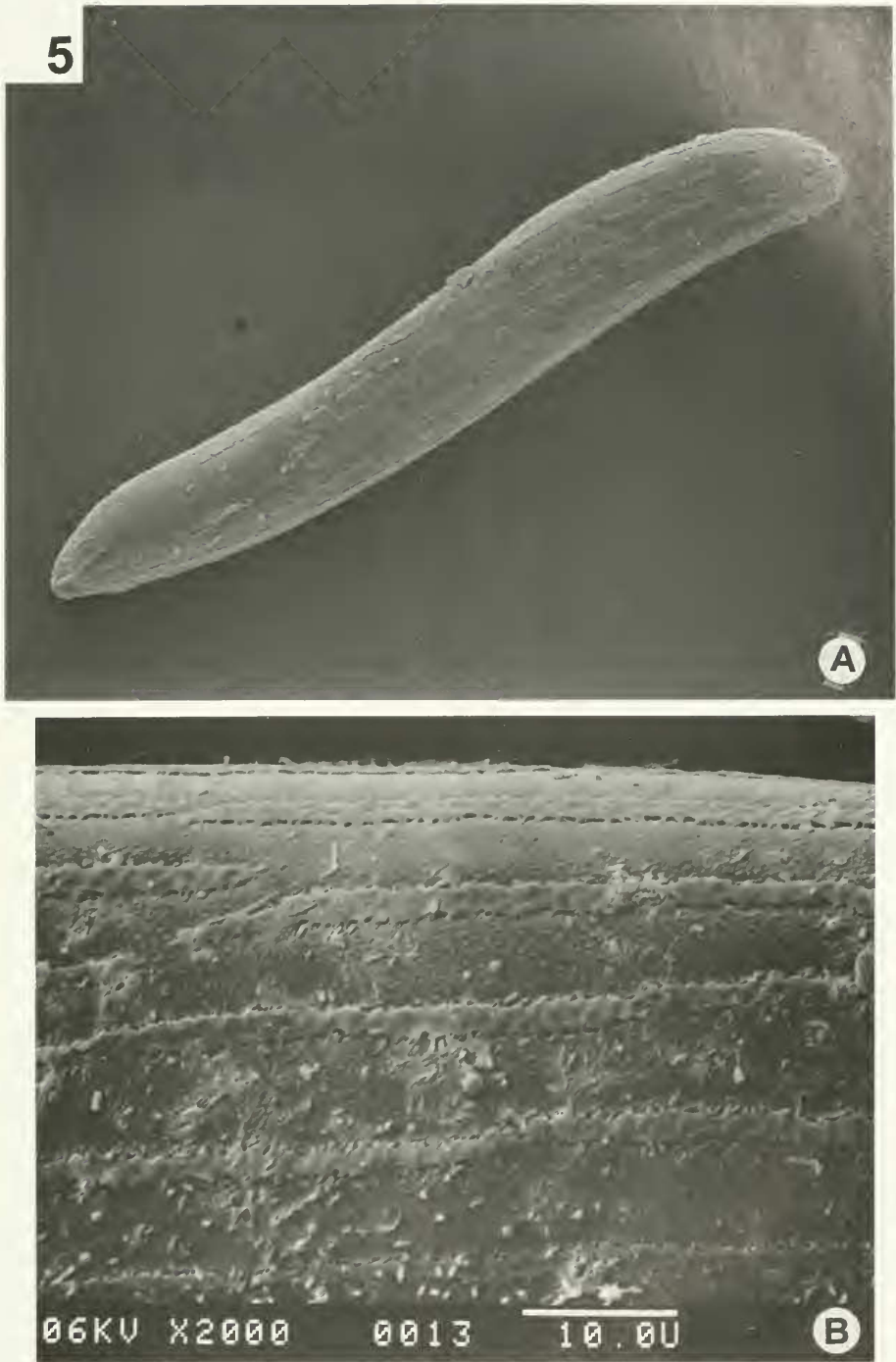


Fig. 5. *Culicoides venustus*, egg. A, Lateral view of entire egg. B, Detail of egg surface.

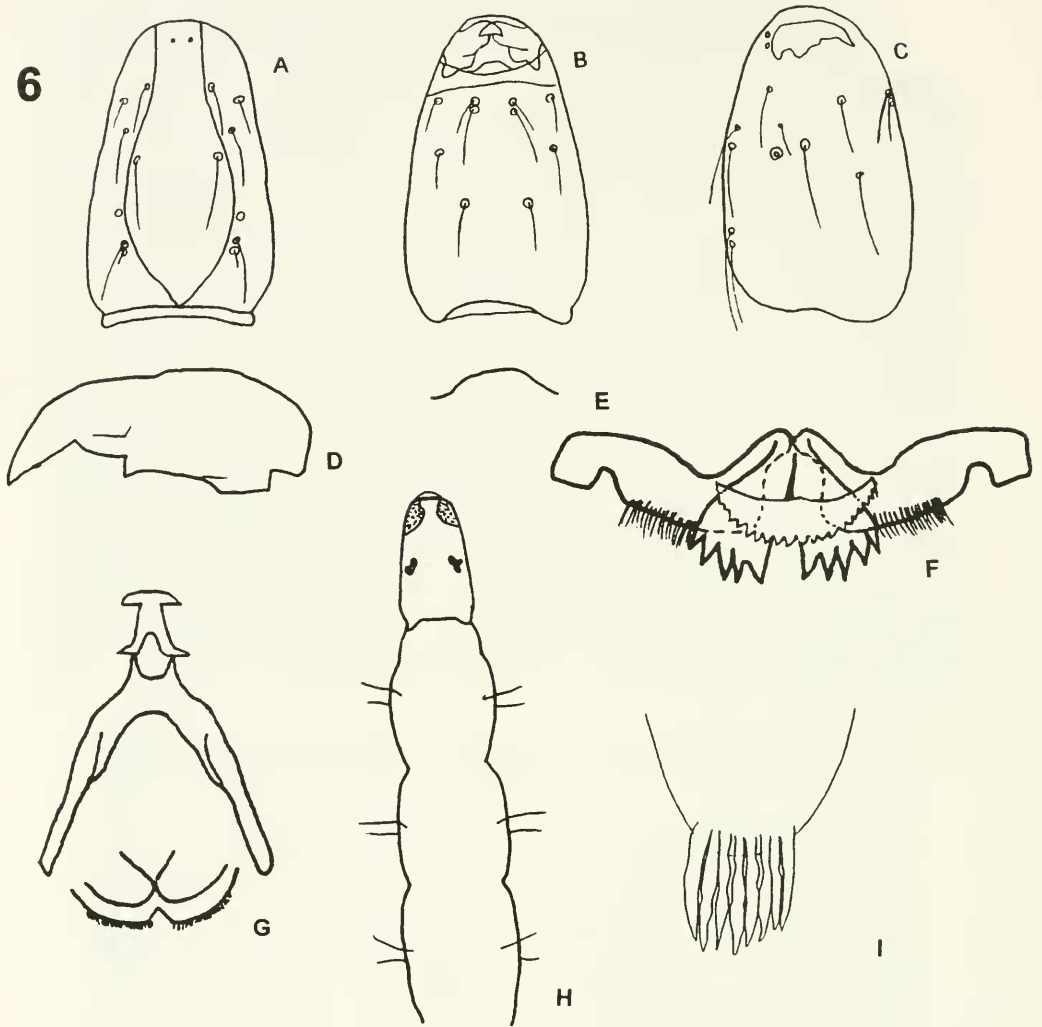


Fig. 6. *Culicoides vetustus*, larva. A-C, Head capsule. A, Dorsal view. B, Ventral view. C, Lateral view. D, Mandible. E, Hypostoma. F, Epipharynx. G, Hypopharynx. H, Head and thorax, dorsal view. I, Anal papillae, caudal segment.

slender bristle, longer and thinner than 1 and 3.

Material examined.—Laboratory-reared from eggs deposited by individual females collected in San Bernardino County, CA, Morongo Valley, 14-V-96 (n = 8); 15-VI-95 A5 (n = 17), paratypes: slides 17-18; 14-V-96 A42 (n = 3), paratypes: slide 1.

Adults.—Female: Wing length 1.24 (1.1-1.4, n = 20) mm. *Head*: Eyes (Fig. 8A) separated by single facet width; without interfacetal hairs. Antenna (Fig. 8B) lengths

of flagellomeres of holotype 46-28-28-30-30-30-33-33-43-46-46-50-65; antennal ratio 0.97 (0.92-1.0, n = 18); well-developed sensilla coeloconica present on flagellomeres 1, 11-13. Palpus (Fig. 8C) with lengths of segments 24-50-78-30-33; third segment swollen, with round moderately deep sensory pit near apex. Palpal ratio 2.06 (1.8-2.3, n = 21); proboscis long, P/H ratio 0.95 (0.81-1.3, n = 19). *Thorax*: Brown, lateral aspects of scutum darker. Legs brown, lacking pattern; hind tibial

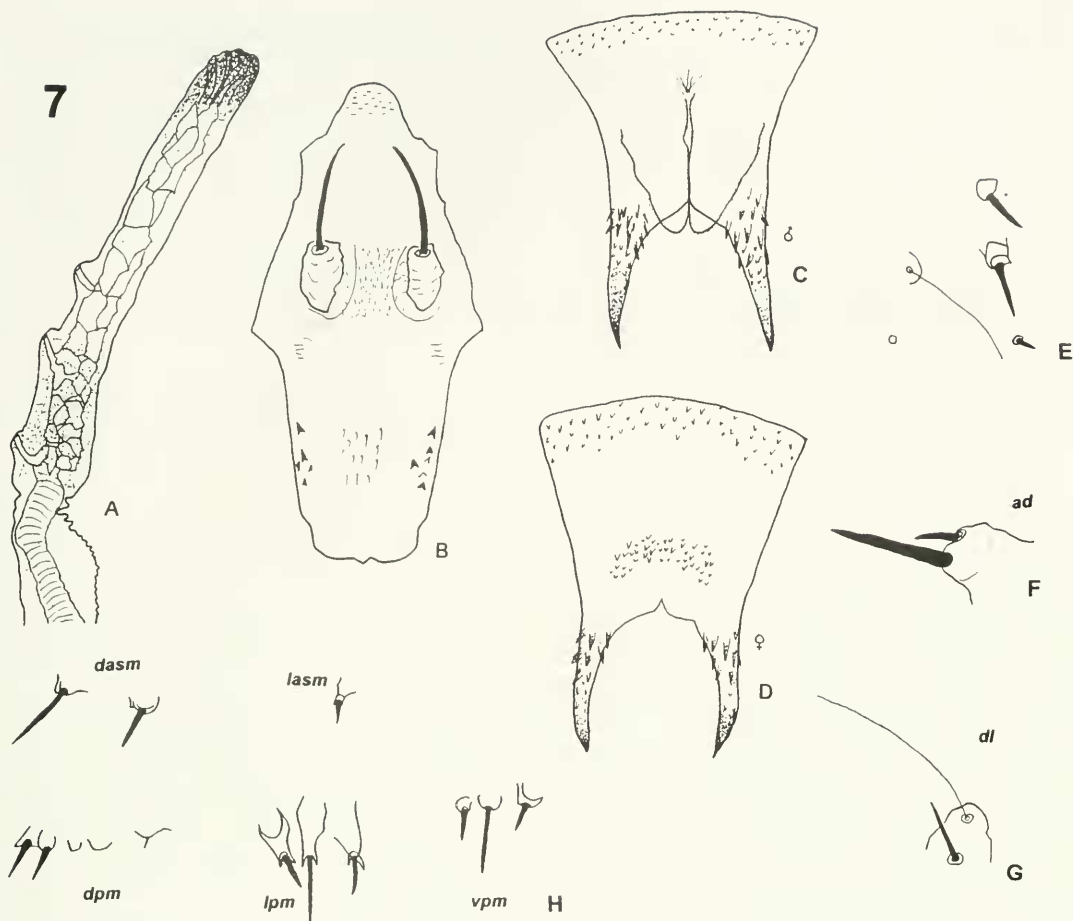


Fig. 7. *Culicoides vetustus*, pupa. A, Respiratory horn. B, Operculum. C, Caudal segment, male. D, Caudal segment, female. E, Dorsal tubercles. F, ad tubercle. G, dl tubercle. H, Abdominal chaetotaxy.

comb (Fig. 8D) with 5 spines, second from spur longest. Wing (Fig. 8E) uniformly grayish with pale spot on anterior margin just past second radial cell. Halter pale. *Abdomen*: Brown. Two slightly unequal spermathecae (Fig. 8F), plus rudimentary third; functional spermathecae with short slender necks; length 44 (35–51, n = 18) μ for larger, 42 (34–50, n = 17) μ for smaller.

Male: Genitalia (Figs. 8G–H): Sternite 9 with broad moderately deep caudomedian excavation, ventral membrane spiculate; tergite 9 with prominent triangular apico-lateral processes, moderately broad, slightly divergent; the caudal margin between them transverse without medial cleft. Gonocoxite moderately stout; ventral root more slender

than dorsal root; gonostylus curved distally, moderately slender, with moderately broad blunt tip. Aedeagus Y-shaped; basal arch extending half of total length; basal arms short, moderately slender, recurved at extreme apex, distal portion moderately broad. Paramere (Fig. 8H) with distinct anterior process; midpoint straight; apex diverging with three sharp medial spines, tip sharply pointed, diverging.

Distribution.—Mojave and Colorado deserts of southern California.

Material examined.—**HOLOTYPE**: ♀, Riverside County, CA, Deep Canyon, nr. Palm Desert, 29-VI-89 (B. A. Mullens), CO₂-baited trap. **Allotype**: ♂, San Bernardino County, CA, Big Morongo Canyon,

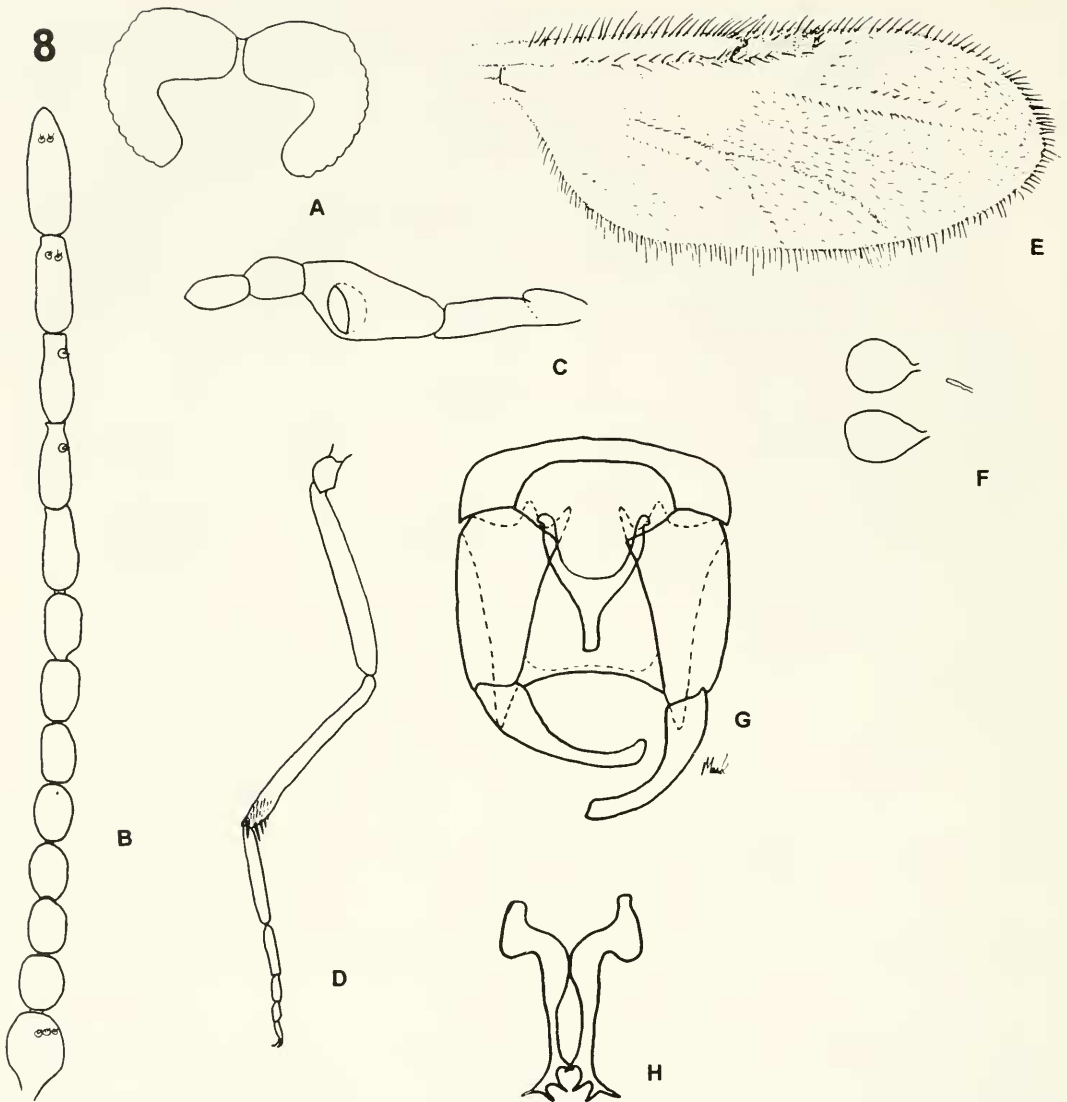


Fig. 8. *Culicoides vetustus*, adult. A-F Female. G-H, Male. A. Eye separation. B. Antenna. C. Palpus. D. Hind leg. E. Wing. F. Spermathecae. G. Genitalia, parameres omitted. H. Parameres.

Morongo Valley, 15-VI-95 (Breidenbaugh), laboratory-reared from wild-collected female. PARATYPES: 1 ♀, same data as allotype; 16 ♀, 11 ♂ same data as allotype, except collected between 14-V-96 and 2-VII-96, some reared from wild-collected ♀; 9 ♀, San Diego County, CA, Anza Borrego State Park, Yaqui Well, 26-IV-97 (Breidenbaugh), CO₂-baited trap.

Biology and rearing.—Adult females collected in CO₂-baited traps readily fed

through an artificial membrane and deposited eggs in the laboratory. Larvae exhibited the slowest development of any species reared on the agar system. Average clutch size was 86 ± 35 eggs ($n = 52$ females); these hatched in 7 ± 1 days with $67 \pm 25\%$ fertility ($n = 25$ females). Average development period to pupation was 117 ± 28 days ($n = 46$). Adults emerged 4 ± 1 days later ($n = 39$).

It is not known if the rearing medium

was responsible for the slow development or whether this is the normal condition for larvae of this species. The percentage of larvae reaching adulthood (approx. 31%) was similar to the other species reared (Breidenbaugh and Mullens, in preparation). Despite extensive observations, larvae were never seen feeding on nematodes; thus, this species was probably utilizing other microorganisms present in the cultures.

Discussion.—As discussed by Blanton and Wirth (1979), adults of a number of Nearctic *Culicoides* do not key readily to the subgenera described by Vargas (1960). This is true for *C. vetustus*. From adult and larval characteristics presented here, we are tentatively placing *C. vetustus* in the *biguttatus* species group. For example, the mandibles of the larvae resemble those illustrated by Murphree and Mullen (1991) of *Culicoides biguttatus* (Coquillett) and *Culicoides spinosus* Root and Hoffman except for the depth of the notch on the subapical margin of the mandible of *C. vetustus*. The number of dorsal-comb teeth (5) on the epipharynx is the same for *C. spinosus*. However, the lateral arms of the epipharynx are notched in *C. vetustus*, but not in *C. spinosus* or *C. biguttatus*. The hypostoma of *C. biguttatus* and *C. vetustus* are very similar.

The pupal operculum of *C. vetustus* resembles that of *C. biguttatus* and *C. spinosus*. However, the operculum of *C. vetustus* has fewer spines, and these are limited to the lateral margins. The respiratory horn of *C. vetustus* is more similar to that of *C. spinosus*, darkened at the tip and with 3 spiracular openings visible.

The range of *C. vetustus* overlaps only with *C. sublettei* Atchley and *C. usingeri* Wirth of the *biguttatus* group. The male genitalia of these species are similar in several respects, including the general shape of the parameres.

The apices of the parameres of *C. vetustus* are divided into 3–4 short spines and the distomedian process of the aedeagus is

bluntly rounded. The aedeagus of *C. vetustus* is more slender than that of *C. sublettei* or *C. usingeri*, and there are 5 tibial spines in *C. vetustus* and *C. sublettei*, but 4 in *C. usingeri*. The females of *C. vetustus* lack a very distinctive wing pattern. However, they can be distinguished clearly from the other species in the *biguttatus* group and from *Culicoides piliferus* Root and Hoffman, which it superficially resembles, by the sensorial pattern (1, 11–13).

Etymology.—Latin *vetustus* for long-lived, referring to the lengthy development period of the larvae.

DISCUSSION

Most descriptions of *Culicoides* species have included only the adult stage or in some cases, only the female. This is a result of the relative difficulty in locating developmental sites or collecting and associating males. The collection of host-seeking females, use of an artificial host, and a laboratory rearing system proved a very successful way of associating immature stages with adults. This approach should be useful for many species whose adults are known. The rearing technique is advantageous in that the entire cohort from an isofemale is unquestionably conspecific. In contrast, field-collected larvae, if they are located, generally must be killed and slide-mounted for identification. Consequently, an incorrect association with other larvae in the collection which have been reared to adults is possible.

The rearing method used here probably results in a narrower range of measurements for morphometric analysis, than if field-collected material were examined. Environmental homogeneity and the genetic similarity of laboratory-reared sibling larvae can result in morphological measurements that are unnaturally similar. The descriptions herein, however, will allow field-collected immatures to be identified and measured to better characterize the range of natural variability in characters such as size.

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